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Melanoderma microcarpum gen. et sp. nov. (*Basidiomycota*) from China

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Abstract — A new genus *Melanoderma* (*Polyporales, Basidiomycota*), collected from Hainan and Hunan Provinces in southern China, is proposed and typified by *M. microcarpum*. The new species is characterized by a perennial habit, pileate to effused-reflexed basidiocarps with a black crust at the pileal surface, small round pores, a dimitic hyphal system with clamped generative hyphae, dextrinoid and cyanophilous skeletal hyphae, cylindrical, thin-walled, non-amyloid, non-dextrinoid and acyanophilous basidiospores, the presence of apically encrusted cystidia, and lack of both hyphal pegs and dendrohyphidia. The new genus is compared with related genera of *Australoporus*, *Datronia*, *Dichomitus*, *Megasporoporia,* and *Perenniporia*. rDNA sequence analysis supports *Melanoderma* as a monophyletic clade that is closely related to *Vanderbylia.*

Key words — polypore, lignicolous fungi, poroid fungi, phylogeny, taxonomy

Introduction

During investigations on lignicolous and poroid fungi in China, three specimens were collected from Hainan and Hunan Provinces in southern China. These collections are microscopically similar to *Perenniporia* Murrill in having a dimitic hyphal system with clamped generative hyphae and dextrinoid and cyanophilous skeletal hyphae, but differ from *Perenniporia* in having thin-walled, non-dextrinoid and acyanophilous basidiospores. These collections may also be confused with species of *Australoporus* P.K. Buchanan & Ryvarden, *Datronia* Donk, *Dichomitus* D.A. Reid and *Megasporoporia* Ryvarden & J.E. Wright, but have distinct differences. Since the species cannot be accommodated in any known genera, we propose a new genus for these collections. To support our proposal as well as to evaluate the position of the new genus and related taxa, phylogenetic analysis was performed using 28S rDNA sequence data.

Materials & methods

Morphological studies. — The studied specimens are deposited at the herbarium of Beijing Forestry University (BJFC) and the herbarium of Institute of Applied Ecology, Chinese Academy of Sciences (IFP). The microscopic procedure follows Cui et al. (2007). In presenting the variation in the size of the spores, 5% of measurements were excluded from each end of the range, and given in parentheses. In the text the following abbreviations are used: IKI = Melzer's reagent, IKI– = negative in Melzer's

Table 1. Species and sequences database accession numbers used in this study.

Species name	Sample no.	GenBank no.
Abundisporus sclerosetosus Decock & Laurence	MUCL 41438	FJ393868
A. violaceus (Wakef.) Ryvarden	MUCL 38617	FJ393867
Antrodia malicola (Berk. & M.A. Curtis) Donk	TFRI 349	EU232296
	FP 104329	EU232297
Antrodiella americana Ryvarden & Gilb.	L 3468	EU232269
	HHB 4100	EU232270
Byssomerulius corium (Pers.) Parmasto	KHL 8593	AY586640
Ceriporia reticulata (Hoffm.) Domański	KHL 11981	EU118614
Donkioporia expansa (Desm.) Kotl. & Pouzar	MUCL 35116	FJ393872
Fomitopsis pinicola (Sw.) P. Karst.	TFRI 513	EU232290
Ganoderma applanatum (Pers.) Pat.	BCRC 36091	EU232274
Gloeocystidiellum aspellum Hjortstam	LIN 625	AF506432
G. porosum (Berk. & M.A. Curtis) Donk	FCUG 2661	AF310099
	FCUG 2768	AF310100
Melanoderma microcarpum	DAI 9811	HQ678175
	DAI 8116	HQ678176
Perenniporia corticola (Corner) Decock	DAI 7330	HQ654108
P. cystidiata Y.C. Dai et al.	CUI 8459	HQ654113
P. detrita (Berk.) Ryvarden	MUCL 42649	FJ393866
P. martia (Berk.) Ryvarden	CUI 7992	HQ654114
	MUCL 41677	FJ393859
	MUCL 41678	FJ393860
P. medulla-panis (Jacq.) Donk	MUCL 43520	FJ393875
	MUCL 49581	FJ393876
P. truncatospora (Lloyd) Ryvarden	CUI 6987	HQ654112
Perenniporiella chaquenia Robledo & Decock	MUCL 47648	FJ393856
	MUCL 49758	FJ393857
Phellinus alni (Bondartsev) Parmasto	TW 162	AF311025
Pyrofomes demidoffii (Lév.) Kotl. & Pouzar	MUCL 41034	FJ393873
Trametes elegans (Spreng.) Fr.	BCC 23750	FJ372713
	BCC 23751	FJ372714
Vanderbylia vicina (Lloyd) D.A. Reid	MUCL 44779	FJ393862
Wrightoporia lenta (Overh. & J. Lowe) Pouzar	KN 150311	AF506489

reagent, KOH = 5% potassium hydroxide, CB = Cotton Blue, CB+ = cyanophilous, CB– = acyanophilous, L = mean spore length (arithmetic average of all spores), W = mean spore width (arithmetic average of all spores), Q = variation in the L/W ratios between the specimens studied, n = number of spores measured from given number of specimens. Sections were studied at magnification up to ×1000 using a Nikon Eclipse E 80i microscope and phase contrast illumination. Drawings were made with the aid of a drawing tube. Special colour terms follow Petersen (1996) and Anonymous (1969).

Molecular procedures and phylogenetic analyses. — DNA was extracted from dried herbarium materials following the methods of Gardes & Bruns (1993) and Cui et al. (2008) with some modifications. In the study, nuclear ribosomal RNA genes were used to determine the phylogenetic position of the new species. Primers for PCR amplification were LROR and LR7 described by White et al. (1990). LSU sequences of the new taxa were deposited in GenBank, and used as references to blast against GenBank, similar sequences were downloaded, taxa used in the phylogenetic analyses are listed in Table 1. Maximum parsimony and Bayesian analysis were applied to the nLSU dataset. All characters were treated as unordered and equal weight. Maximum parsimony analysis (PAUP* version 4.0) was used (Swofford 2002), based on strict heuristic searches with a tree-bisection reconnected (TBR) branch swapping algorithm, stepwise addition and collapse of branch if maximum length is zero. Bootstrap values were calculated from 1000 replicates. Bayesian analysis with MrBayes3.1.2 (Ronquist & Huelsenbeck 2003) implementing the Markov Chain Monto Carlo (MCMC) technique and parameters predetermined with MrMODELTEST2.3 was performed. When four simultaneous Markov chains were run with 100,000 generations, the average standard deviation of split frequencies was 0.00849.

Taxonomy

Melanoderma B.K. Cui & Y.C. Dai, **gen. nov.**

MycoBank MB 519872

Carpophorum perenne, pileatum vel effuso-reflexum, pileatum nigrum, crustum. Contextum cremea bubalina; facies pororum nivea vel cremea, pori rotundi. Systema hypharum dimiticum, hyphae generatoriae fibulatae, hyphae skeletales dextrinoideae; cystidia pallida, ventricosa, incrustata. Sporae hyalinae, cylindricae, IKI–, CB–.

Type Species: *Melanoderma microcarpum* B.K. Cui & Y.C. Dai

Etymology: from Greek *melano-* (= black), *derma* (= skin); referring to the black crust at pileal surface.

Basidiocarps perennial, pileate to effused-reflexed, pileus circular to irregularly formed. Pileal surface black when fresh, colour unchanged when dry, concentrically zonate, glabrous; margin obtuse. Pore surface white when fresh, cream buff when dry; pores circular, dissepiments thick, entire. Context cream-buff, woody hard. Tubes cream-buff, woody hard, stratified. Hyphal system dimitic; generative hyphae bearing clamp connections; skeletal hyphae dextrinoid, CB+; tissue unchanged in KOH. Generative hyphae clamped, scanty, hyaline, thin-walled, unbranched; skeletal hyphae dominant, thick-walled to subsolid, usually branched, strongly interwoven. Cystidia clavate to ventricose,

hyaline, thin-walled, usually apically encrusted; cystidioles clavate, hyaline, thin-walled. Basidia clavate, with four sterigmata and a basal clamp connection; basidioles similar in shape to basidia, but slightly smaller. Rhomboid crystals frequently present in trama and hymenium. Basidiospores cylindrical, hyaline, thin-walled, smooth, IKI–, CB–.

The genus *Melanoderma* is characterized by a perennial growth habit, pileate to effused-reflexed basidiocarps with a black crust at pileal surface, a white to cream-buff pore surface, and small, round pores. A dimitic hyphal system with clamped generative hyphae, dextrinoid and cyanophilous skeletal hyphae, cylindrical, thin-walled, non-amyloid, non-dextrinoid and acyanophilous basidiospores, apically encrusted cystidia, and lack of hyphal pegs and dendrohyphidia also distinguish the genus.

Melanoderma microcarpum B.K. Cui & Y.C. Dai, **sp. nov.** Fig. 1

MycoBank MB 519873

Carpophorum perenne, pileatum vel effuso-reflexum. Contextum cremea bubalina; facies pororum nivea vel cremea, pori rotundi, 7–9 per mm. Systema hypharum dimiticum, hyphae generatoriae fibulatae, hyphae skeletales dextrinoideae, hyphae skeletales contexti 1.5–5.7 µm; cystidia frequentia, pallida, ventricosa, incrustata. Sporae hyalinae, cylindricae, IKI–, CB–, 5.1–6.4 × 1.9–2.7 µm.

Type: China. Hunan Province, Yizhang County, Mangshan Forest park, on fallen angiosperm trunk, 25.VI.2007 *Dai 8116* (**holotype** in BJFC; **isotype** in IFP).

Etymology: from Greek *micro-* (= small), *-carpus* (= fruited); referring to the small basidiocarps.

Fruitbody — Basidiocarps perennial, pileate to effuse-reflexed, narrowly attached, woody hard upon drying, without odour or taste, pileus circular to irregularly formed, projecting up to 1.6 cm, 1.5 cm broad and 5 mm thick at base. Pileal surface black when fresh and dry, concentrically zonate, glabrous; margin obtuse, cream-buff when juvenile, becoming black when mature. Pore surface white when fresh, cream to cream-buff when dry; pores distinctly circular, 7–9 per mm, dissepiments thick, entire. Context cream-buff, woody hard, up to 1 mm thick, upper surface with a black crust. Tubes cream-buff, woody hard, stratified, about 1mm long in each layer.

Hyphal structure — Hyphal system dimitic; generative hyphae bearing clamp connections; skeletal hyphae dextrinoid, CB+; tissue unchanged in KOH.

Context — Generative hyphae scanty, hyaline, thin-walled, rarely branched, 1.5–3.6 µm in diam; skeletal hyphae dominant, hyaline, thick-walled to subsolid, often branched, interwoven, 1.5–5.7 µm in diam.

Tubes — Generative hyphae scanty, hyaline, thin-walled, unbranched, 1.2–3 µm in diam; skeletal hyphae dominant, thick-walled to subsolid, usually branched, strongly interwoven, 1.2–5 µm in diam. Cystidia clavate to ventricose,

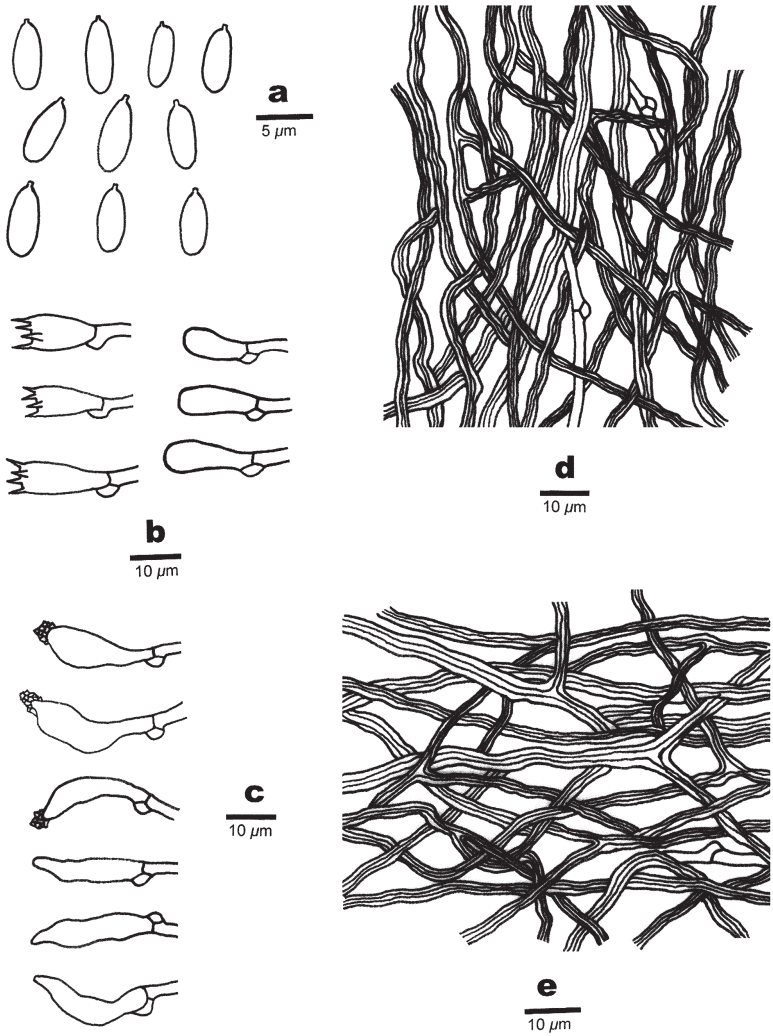

Fig. 1. Microscopic structures of *Melanoderma microcarpum* (drawn from the holotype). a: Basidiospores. b: Basidia and basidioles. c: Cystidia and cystidioles. d: Hyphae from tubes. e: Hyphae from context.

hyaline, thin-walled, usually apically encrusted, 20–32 × 4.8–7.6 μm; cystidioles clavate, hyaline, thin-walled, 17.6–24 × 4–6.2 μm. Basidia clavate, with four sterigmata and a basal clamp connection, 9.8–16 × 5–6.5 μm; basidioles similar

in shape to basidia, but slightly smaller. Rhomboid crystals frequently present in trama and hymenium.

Spores — Basidiospores cylindrical, hyaline, thin-walled, smooth, IKI–, CB–, (5–)5.1–6.4(–7.2) × (1.8–)1.9–2.7(–3) μm, L = 5.92 μm, W = 2.28 μm, Q = 2.25–2.79 (n = 90/3).

Type of rot — White rot.

Additional specimens examined: **CHINA. Hai Province**, Lingshui County, Diaoluoshan Forest Park, on fallen angiosperm trunk, 29.V.2008 *Dai 9811* (BJFC & IFP). Ledong County, Jianfengling Nature Reserve, on fallen angiosperm branch, 11.V.2009 *Cui 6582* (BJFC & IFP).

Phylogeny

A primarily phylogenetic analysis on the position of the new species was done, and the parsimony consensus tree (Fig. 3) was made up based on the 28S rDNA sequences, in the tree, sequences of *Melanoderma microcarpum* were grouped together as a monophyletic cluster with strong support by Bayesian analysis and Maximum Parsimony analysis.

Discussion

Melanoderma microcarpum is characterized by a perennial growth habit, pileate to effuse-reflexed basidiocarps with a distinct black crust at pileal surface, small and round pores, dimitic hyphal system with clamped generative hyphae, dextrinoid and cyanophilous skeletal hyphae, cylindrical, thin-walled, non-amyloid, non-dextrinoid and acyanophilous basidiospores, presence of apically encrusted cystidia, and lack of both hyphal pegs and dendrohyphidia.

Melanoderma microcarpum has dextrinoid and cyanophilous skeletal hyphae, which may be confused with species of *Perenniporia*. However, *Perenniporia* species have thick-walled, cyanophilous basidiospores (Dai et al. 2002), while *M. microcarpum* has thin-walled, acyanophilous basidiospores.

Melanoderma microcarpum is similar to *Megasporoporia* in sharing a dimitic hyphal system with clamped generative hyphae, dextrinoid and cyanophilous skeletal hyphae, cylindrical and thin-walled basidiospores, and rhomboid crystals in trama and hymenium. *Megasporoporia* species, however, usually have resupinate basidiocarps with big pores, large basidiospores, numerous hyphal pegs and dendrohyphidia, lack cystidia (Ryvarden et al. 1982, Dai & Wu 2004, Dai & Cui 2008, Du & Cui 2009). *Melanoderma microcarpum* has small pores (7–9 per mm), relatively small basidiospores, apically encrusted cystidia, and lacks hyphal pegs and dendrohyphidia. Furthermore, it has pileate basidiocarps with a black crust at the upper surface.

Australoporus has a dimitic hyphal system, dextrinoid skeletal hyphae, apically encrusted cystidia and cylindrical and thin-walled basidiospores, similar to features found in *Melanoderma,* but *Australoporus* has an ungulate basidiocarp without crust at pileal surface, pale pink to orange pore surface,

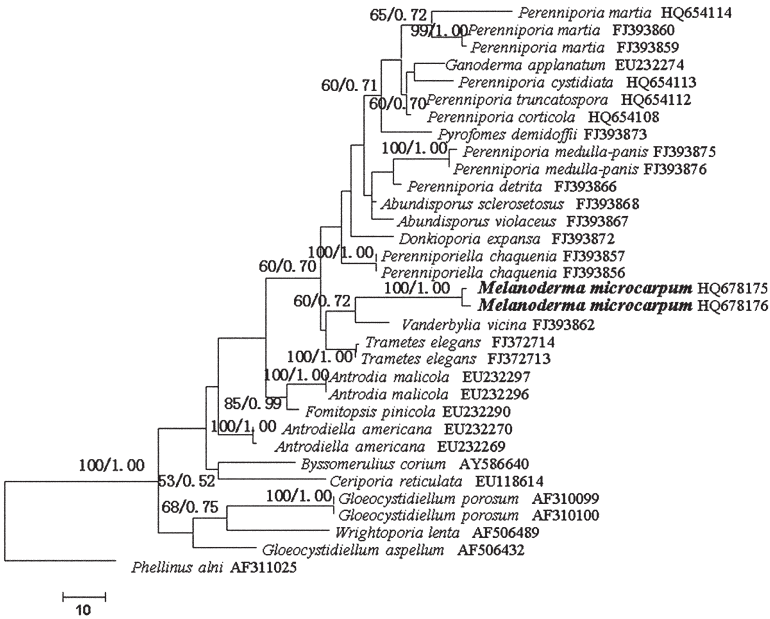

Fig. 2. Strict consensus tree illustrating the phylogeny of *Melanoderma microcarpum* generated by Bayesian and Maximum Parsimony analyses based on 28S rDNA sequences. Posterior probabilities and bootstrap values (≥50%) are shown the above or below branches.

and finely encrusted skeletal hyphae in dissepiments (Buchanan & Ryvarden 1988, Ryvarden 1991).

Datronia and *Dichomitus* have a dimitic hyphal system with clamped generative hyphae, cyanophilous skeletal hyphae, and cylindrical and thin-walled basidiospores, but those genera differ from *Melanoderma* in having non-dextrinoid skeletal hyphae and lacking apically encrusted cystidia (Masuka & Ryvarden 1999; Ryvarden 1991; Núñez & Ryvarden 2001).

The phylogenetic analysis confirmed *Melanoderma* as a distinct monophyletic cluster closely related to the genus *Vanderbylia* D.A. Reid, which, however, has distinctly thick-walled, dextrinoid, and cyanophilous basidiospores (Decock & Masuka 2003). Based on both morphological and molecular data, a new genus is proposed for the new species.

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